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Absence of Age and Gender Effects on the Pharmacokinetics of a Single 500-Milligram Oral Dose of Levofloxacin in Healthy Subjects

S. C. CHIEN, A. T. CHOW,* J. NATARAJAN, R. R. WILLIAMS, F. A. WONG, M. C. ROGGE, AND R. K. NAYAK

The R. W. Johnson Pharmaceutical Research Institute, Raritan, New Jersey 08869

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The influence of age and gender on the pharmacokinetics of levofloxacin in healthy subjects receiving a single oral 500-mg dose of levofloxacin was investigated in this parallel design study. Six young males (aged 18 to 40 years), six elderly males (aged ≥65 years), six young females (aged 18 to 40 years), and six elderly females (aged ≥65 years) were enrolled and completed the study. The study reveals that the bioavailability (rate and extent) of levofloxacin was not affected by either age or gender. In both age (young and elderly) and gender (male and female) groups of subjects, peak concentrations in plasma were reached at approximately 1.5 h after dosing; renal clearance of levofloxacin accounted for approximately 77% of total body clearance, and approximately 76% of the administered dose was recovered unchanged in urine over the 36 h of collection. The apparent differences in the calculated pharmacokinetic parameters for levofloxacin between the age groups (young versus elderly) and between the gender groups (males versus females) could be explained by differences in renal function among the subjects. A single dose of 500 mg of levofloxacin administered orally to both young and old, male and female healthy subjects was found to be safe and well tolerated. As the differences in levofloxacin kinetics between the young and the elderly or the males and the females are limited and are mainly related to the renal function of the subjects, dose adjustment based on age or gender alone is not necessary.

Levofloxacin, (−)-(*S*)-9-fluoro-2,3-dihydro-3-methyl-10-(4-methyl-1-piperazinyl)-7-oxo-7H-pyrido[1,2,3-de][1,4]benzoxazine-6-carboxylic acid hemihydrate, a fluoroquinolone antibacterial agent, is the active (−)-*S*-isomer of the racemic drug substance ofloxacin. The pharmacokinetic profiles of levofloxacin and ofloxacin are very similar (7). Levofloxacin was approved for marketing in the United States in December 1996.

The pharmacokinetics of many drugs are altered by age (14) and/or by gender (9). Molinaro et al. (6) reported that, compared with healthy young subjects, the elimination of ofloxacin was delayed in elderly patients. The objective of this study was to evaluate the influence of age and gender on the pharmacokinetics of levofloxacin in healthy subjects.

(Part of the study results were presented at the 34th Interscience Conference on Antimicrobial Agents and Chemotherapy, 4 to 7 October 1994, in Orlando, Fla. [7a], and at the 5th International Symposium on New Quinolones, 1994, in Singapore.)

MATERIALS AND METHODS

Subjects. Twenty-four healthy male and female volunteer subjects entered the study. Participants were stratified first by gender and then by age (either 18 to 40 or ≥65 years). Qualified subjects had to have normal findings in the prestudy medical history and physical examination performed within 2 weeks of the study entry. Subject eligibility was restricted to those with no evidence of clinically significant hematologic, serum chemistry, or urinalysis laboratory values. All subjects were required to have a calculated creatinine clearance (CL_{CR}) greater than 20 ml/min as determined by the method of Cockcroft and Gault (2). Female subjects must have been postmenopausal (naturally or surgically induced) for at least 1 year or using acceptable methods of contraception. Key exclusion criteria included any history of a significant gastrointestinal condition that could interfere with the absorption or disposition of the study medication, a previous history of allergy to a fluoroquinolone, alcohol or controlled substance abuse, or use of an investigational agent within 30 days of study entry. Potential subjects were also excluded if they used any medication within 3 days prior to administration of the first study dose or suffered from an acute illness within 1 week of study entry. Female subjects were excluded if they had a positive pregnancy test. All subjects signed an informed consent form approved by the investigational review board.

Study design. This phase I study was conducted as a single-center, parallel group design. Subjects were administered a single oral tablet consisting of 500 mg of anhydrous levofloxacin (The R. W. Johnson Pharmaceutical Research Institute). The study drug was administered with 8 oz of water. Subjects fasted for at least 8 h prior to and through 2 h after dosing; water was allowed ad libitum except 2 h before and through 2 h after dosing. A standard breakfast was served 2 h after the dose. Ingestion of alcohol was not permitted within 48 h prior to study entry and during the study period. Smoking was not allowed during the course of the study. Subjects were confined from the evening prior to administration of the single dose until after all blood samples had been collected, approximately 36 h postdose.

Sample collection. Samples (5 ml) of venous blood for determination of plasma levofloxacin concentrations were collected from an indwelling catheter following the single-dose treatment immediately prior to the dose and then at the following times postdose: 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 4.0, 6.0, 8.0, 12.0, 24.0, and 36.0 h. Blood samples were collected in heparinized tubes and centrifuged; the plasma was separated and stored at −20°C until assayed. Urine samples were collected during the following time periods: 0 (predose), 0 to 4, 4 to 8, 8 to 12, 12 to 24, and 24 to 36 h postdose.

Safety analysis. Safety evaluations were based on changes in physical examination findings, vital signs, and clinical laboratory tests (hematology, blood chemistry, and urinalysis) from predosing to postdosing and adverse events reported throughout the study. Subjects were observed for 36 h after dosing.

Analytical procedures. Plasma and urine levofloxacin concentrations were determined by a sensitive and specific reversed-phase high-performance liquid chromatography assay with UV detection (10). Briefly, the procedure utilized a single liquid-liquid extraction step with methylene chloride as the extraction solvent and ciprofloxacin as the internal standard. The chromatographic separation of levofloxacin from endogenous substances was accomplished on a 5-μm ODS-2 (4.6 by 150-mm) reversed-phase column. Elution was performed isocratically with a mixture of 0.005 M copper(II) sulfate pentahydrate in 0.01 M *l*-isoleucine and methanol (87.5:12.5 [vol/vol]) at a flow rate of 1.0 ml/min. UV detection was accomplished at 330 nm. The assay was validated from 80 to 5,120 ng/ml for plasma and 25 to 2,000 μg/ml for urine. Quality control samples were analyzed along with the study samples. The mean variability of the standards for the plasma assay was 3.02% with a maximum variability of 5.22% at 80 ng/ml. The mean variability of the standards for the urine assay was 4.59% with a

* Corresponding author. Mailing address: The R. W. Johnson Pharmaceutical Research Institute, 1000 Route 202, Raritan, NJ 08869-0602. Phone: (908) 704-4057. Fax: (908) 253-0448.

TABLE 1. Demographic and baseline characteristics

Characteristic	Young males (18–40 yr) (n = 6)	Elderly males (≥65 yr) (n = 6)	Young females (18–40 yr) (n = 6)	Elderly females (≥65 yr) (n = 6)
Race (no. of subjects)				
Caucasian	1	6	3	6
Black	3	0	3	0
Hispanic	2	0	0	0
Age (yr)				
Mean	29.5	69.0	25.3	71.3
Range	22–36	66–75	18–29	66–80
Body wt (kg)				
Mean	77.1	84.7	70.7	60.0
Range	61–88	71–97	51–82	50–71
Serum creatinine (mg/dl)				
Mean	1.18	1.28	1.05	0.97
Range	1.1–1.4	1.1–1.6	0.9–1.3	0.9–1.1
CL_{CR}[a]				
Mean	99.3	65.4	93.8	50.8
Range	90–117	47–80	81–112	39–61

[a] Estimated according to subject's serum creatinine concentration, body weight, age, and gender (2).

maximum variability of 6.57% at 50 μg/ml. Overall, the assay precision (percentage of coefficient of variation) was less than 10%, and the assay accuracy (percentage of deviation from theoretical value) was within 10% of the theoretical values.

Pharmacokinetic analysis. Pharmacokinetic parameters estimated included the area under the plasma concentration-time curve (AUC_{0-t}) as measured by the trapezoidal summation method; $AUC_{0-\infty}$ calculated as $AUC_{0-last} + Cp_{last}/K_e$, where Cp_{last} is the last measurable plasma concentration and K_e is the terminal elimination rate constant calculated as the slope of the terminal log-linear phase of the plasma concentration-time profile; mean residence time calculated as ($AUMC_{0-\infty}/AUC_{0-\infty}$); apparent total body clearance (CL/F) calculated as dose/$AUC_{0-\infty}$ and normalized for body weight; renal clearance (CL_R) calculated as Au/AUC; and, assuming the absorption rate was zero order and absorption was completed at peak time (T_{max}), the apparent volume of distribution (V_d/F) calculated as CL/F · (mean residence time − $T_{max}/2$) and normalized for body weight. AUMC and Au refer to the area under the first moment of the plasma concentration-time curve and the total urinary recovery of levofloxacin at the corresponding AUC intervals, respectively. The peak concentrations of drug in plasma (C_{max}) and the time to reach C_{max} (T_{max}) were estimated by visual inspection of the plasma drug concentration (Cp)-versus-time data.

Statistical analysis. Analysis-of-variance models were used to study the effects of age and gender on the pharmacokinetic parameters (C_{max}, T_{max}, $AUC_{0-\infty}$, V_d/F, terminal plasma elimination half-life [$t_{1/2}$], CL/F, and CL_R). The analysis-of-variance model was based on a 2 × 2 factorial design and included terms for the two main effects (age and gender) and the age group by gender interaction term. If the interaction term was found not to be significant, the model was refitted to the data without the interaction term and the tests for the main effects were carried out. If the interaction term was significant, no further tests for the main effects were carried out. All tests were performed at a 5% level of significance.

The pharmacokinetic parameters (C_{max}, $AUC_{0-\infty}$, CL/F, and CL_R) were further analyzed by taking into consideration the subject's prestudy creatinine clearance values (CL_{CR}). Analysis-of-variance models were fitted to the total body clearance and renal clearance data with the creatinine clearance as a covariate and age group, gender, and age group by gender interaction as factors. Analysis-of-variance models were fitted to the AUC and C_{max} data with the inverse of the creatinine clearance as a covariate and age group, gender, and age group by gender interaction as factors. If the interaction term was found not to be significant, the model was refitted to the data without the interaction term and the tests for the main effects were carried out. If the interaction term was significant, no further tests for the main effects were carried out. All tests were performed at a 5% level of significance. The statistical analysis was carried out with SAS version 6.08 (11).

RESULTS

Demographic and baseline characteristics. Six young males, six young females, six elderly males, and six elderly females participated in and completed the study. The demographic and baseline characteristics are presented in Table 1.

Discontinuation-completion information. All 24 subjects completed the study and were included in the pharmacokinetic and safety analyses.

Pharmacokinetic results. Mean (± standard deviation [SD]) plasma concentration-time curves of levofloxacin in the age group (young or elderly) and in the gender group (males or females) are shown in Fig. 1 and 2, respectively. The mean (± SD) pharmacokinetic parameters determined from this study are summarized in Table 2. Data are grouped according to the gender and the age of the subjects.

In all four groups of subjects, mean peak concentrations in plasma were reached at approximately 1.5 h after dosing; renal clearance of levofloxacin accounted for approximately 77% of total body clearance, and approximately 76% of the dose was recovered in urine over the 36 h of collection.

Analysis-of-variance modeling was used to determine the effects of age and gender on the pharmacokinetics of levofloxacin. Since the interaction term (age group by gender) was found to be not significant, the model was refitted to the data without the interaction term and the tests for the main effects were carried out.

Statistically significant differences in C_{max}, V_d/F, $AUC_{0-\infty}$, $t_{1/2}$, CL/F, and CL_R between the young and the elderly were observed. The differences in Au and T_{max} between the young and the elderly were not significant.

Statistically significant differences in C_{max}, T_{max}, V_d/F, $t_{1/2}$, and CL/F between males and females were observed. The differences in Au, $AUC_{0-\infty}$, and CL_R between males and females were not significant. Although T_{max} was significantly different between males and females, the difference (Δ = 0.5 h) may be an artifact of the sample collection schedule and is clinically irrelevant.

As levofloxacin undergoes limited metabolism and is elimi-

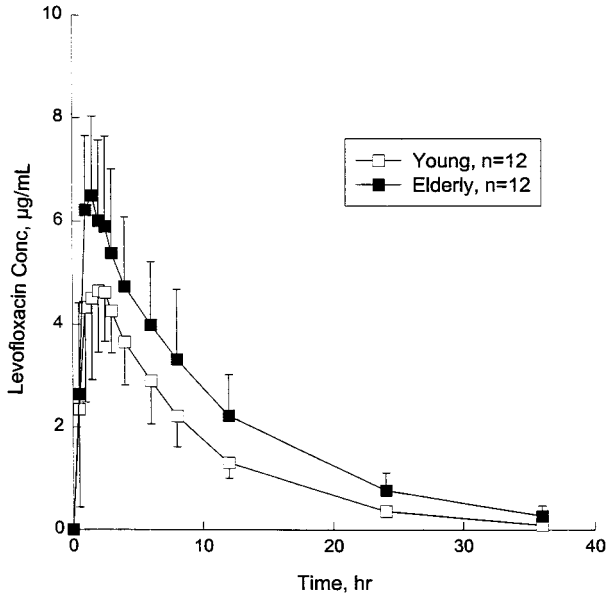

FIG. 1. Mean (± SD) plasma levofloxacin concentrations in the age groups (young and elderly) after a single oral dose of levofloxacin (500-mg tablet).

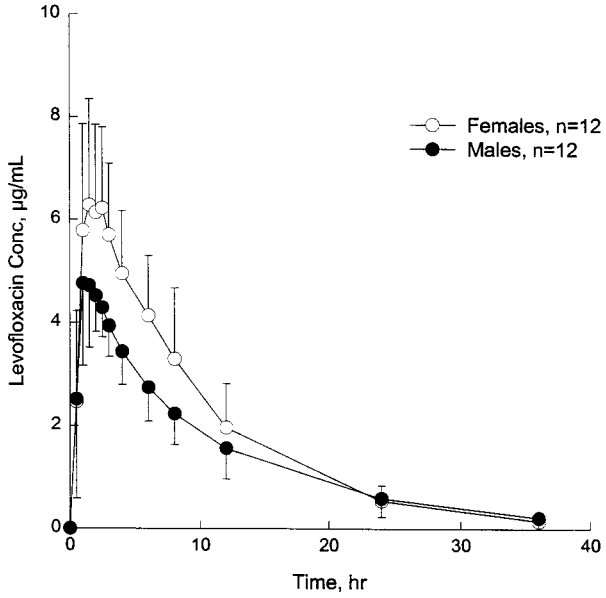

FIG. 2. Mean (± SD) plasma levofloxacin concentrations in the gender groups (males and females) after a single oral dose of levofloxacin (500-mg tablet).

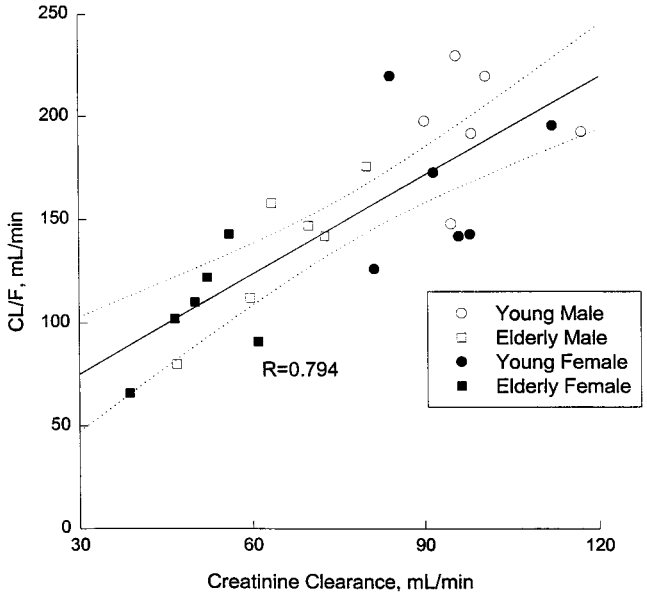

FIG. 3. Total body clearance (CL/F) of levofloxacin versus subject's creatinine clearance (CL_{CR}) for 24 healthy subjects (young males, elderly males, young females, and elderly females) receiving a single oral dose of levofloxacin (500-mg tablet).

nated primarily through the kidney (6), the correlation of the subject's renal function with the clearance of levofloxacin was examined. Calculated creatinine clearances (CL_{CR}) (2) were used as the index of the subject's renal function. As shown in Fig. 3, good correlation was observed between CL_{CR} and CL/F ($r = 0.794$). In addition, high correlations of CL_{CR} with C_{max} ($r = 0.657$) and with $AUC_{0-\infty}$ ($r = 0.786$) were also observed. $t_{1/2}$ was not correlated with CL_{CR} ($r = 0.397$). With respect to the subject's body weight, although it correlated well with C_{max} ($r = 0.714$) and V_d/F ($r = 0.764$), it correlated poorly with $t_{1/2}$ ($r = 0.395$) and CL/F ($r = 0.454$) of levofloxacin.

The differences in the pharmacokinetics (C_{max}, $AUC_{0-\infty}$, CL/F, and CL_R) of levofloxacin between the young and the elderly and between males and females became statistically not significant when the subject's renal function (as indicated by creatinine clearance [CL_{CR}]) was included as a covariate in the analysis-of-variance model. The adjusted means of pharmacokinetic parameters after correction for the subject's renal function (CL_{CR}) are summarized in Table 3.

Safety results. Six of the 24 subjects reported a single adverse event during the study. Three of the subjects reporting adverse events were young women (headache, nausea, and pharyngitis), and one subject in each of three remaining groups reported an adverse event (headache, laceration and abrasion, and hematuria). There were no deaths or serious adverse events during the study, nor did any adverse event require discontinuation. There were no clinically meaningful changes from baseline in any laboratory parameter, vital sign, or physical examination. A single oral dose of 500 mg of levofloxacin administered to both young and old, men and women, was found to be safe. The incidence of adverse events appeared to be similar among the four groups, and an examination of laboratory parameters, physical examinations, and vital signs did not reveal any clinically significant changes after dosing.

TABLE 2. Summary of levofloxacin pharmacokinetic parameters (mean ± SD)

Parameter	Males[a] ($n = 12$)	Females[b] ($n = 12$)	Young[c] ($n = 12$)	Elderly[d] ($n = 12$)
C_{max} (µg/ml)	5.52 ± 1.07	6.96 ± 1.57	5.52 ± 1.02	6.96 ± 1.60
T_{max} (h)	1.2 ± 0.4	1.7 ± 0.5	1.5 ± 0.6	1.4 ± 0.5
V_d/F (liter/kg)	1.11 ± 0.19	0.94 ± 0.14	1.13 ± 0.18	0.92 ± 0.12
$AUC_{0-\infty}$ (µg · h/ml)	54.4 ± 18.9	67.7 ± 24.2	47.5 ± 9.8	74.7 ± 23.3
$t_{1/2}$ (h)	7.5 ± 2.1	6.1 ± 0.8	6.0 ± 0.9	7.6 ± 2.0
Au (% dose [0–36 h])	75 ± 14	77 ± 7	77 ± 10	75 ± 11
CL/F (ml/min)	166 ± 44	136 ± 44	182 ± 35	121 ± 33
CL/F (ml/min/kg)[e]	2.10 ± 0.69	2.07 ± 0.49	2.49 ± 0.47	1.68 ± 0.37
CL_R (ml/min)	126 ± 38	106 ± 40	140 ± 33	91 ± 29

[a] Males (young and elderly).
[b] Females (young and elderly).
[c] Young (males and females); age, 18 to 36 years.
[d] Elderly (males and females); age, 66 to 80 years.
[e] Apparent total body clearance per kilogram of body weight.

DISCUSSION

In past years, the pharmacokinetics of a new agent were often studied only with young healthy male volunteers. Because it is anticipated that levofloxacin will be prescribed for both men and women, as well as elderly and young adult patients, more complete pharmacokinetic studies are needed. Additionally, these studies are warranted because recent find-

TABLE 3. Summary of adjusted means of levofloxacin pharmacokinetic parameters[a]

Parameter	Males	Females	Young	Elderly
C_{max}	5.77	6.71	6.26	6.22
$AUC_{0-\infty}$	60.6	61.6	66.0	56.2
CL/F	160	143	157	146
CL_R	120	112	117	115

[a] The adjusted means were obtained as the predicted values of the pharmacokinetic parameters corresponding to an average creatinine clearance value.

ings have suggested that the volume of distribution and total body clearance of a number of fluoroquinolones including ofloxacin might be affected by age and/or gender (1, 4, 6, 12, 13). For ofloxacin, following multiple 200-mg doses of ofloxacin administered every 12 h, significant accumulation was noted among elderly patients, whereas C_{max}, AUC, and elimination half-life did not increase in younger patients following 7 days of therapy. The most obvious reason for these observed differences is the fact that glomerular filtration diminishes with age (3, 5, 14). Because levofloxacin has a pharmacokinetic profile similar to that of ofloxacin, these findings may also be applicable to this new fluoroquinolone.

As shown in the present study, T_{max} and Au were consistent among the age groups (young and elderly) and gender groups (males and females), indicating that the bioavailability (rate and extent) of levofloxacin was not affected by either age or gender. The apparent volume of distribution of levofloxacin in females is approximately 15% lower than that in males, and in elderly subjects it is approximately 18% lower than that in young adult subjects. As levofloxacin is extensively distributed throughout the body, the above findings are consistent with the fact that lean body mass decreases (8) with advancing age, and that females usually have lower body weight and a higher percentage of body weight as fat than do males. Although small apparent differences in levofloxacin pharmacokinetic parameters were observed between young and elderly subjects and between males and females, these differences were explained by the differences in renal function among the subjects. Dose adjustment based on gender or age alone is not necessary.

Levofloxacin administered orally to both young and old, male and female healthy subjects was found to be safe and well tolerated. The incidence of adverse events was similar among the four groups. This observation was consistent with the analysis of the entire adverse event data in the New Drug Application submission for levofloxacin, in which no significant difference between the sexes was found. Examination of laboratory parameters, physical examinations, and vital signs did not reveal any clinically significant changes after dosing. As the differences in levofloxacin kinetics between the young and elderly, or males and females, are limited and are mainly related to the renal function of the subjects, dose adjustments based on age or gender alone are not necessary.

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